8th annual meeting of the European Society for Molecular Imaging – ESMI
European Molecular Imaging Meeting – EMIM 2013
26 to 28 May, 2013 | Torino Incontra - Centro Congressi | Via Nino Costa 8 | Torino, Italy

PO 09 | Cancer Imaging III
Type: Poster Session
Chair: Uwe Himmelreich (Leuven), Paul van Bergen En Henegouwen (Utrecht)
Date: May 28, 2013
Time: 9:00 - 10:30

Separation of Fluorescent Spectra of quantum dots in Multispectral Imaging Based on Linear Unmixing Vs Non-negative Matrix Factorization in Fluorescence reflectance imaging within Tissue Equivalent Phantom (#308)

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Introduction

Fluorescence reflectance imaging (FRI) is widely used in small animals to record their biological operations and feedbacks. In FRI, proteins or cells are labeled by fluorophores to render cell parts visible. In this study, linear unmixing algorithm have been presented to segregate radiated signals by the FRI method. This method determines the volume distribution of fluorescent proteins in different textures based on optical tomography.

Methods

Optical set up has been designed and executed to make multispectral images containing two filtering sets (in range of 550 to 900 nm) using an EMCCD camera and three laser beams. In this set up, the quantum point sources in tissue equivalent phantom were irradiated by laser beams. After each fluorescence emitted by fluorophores was subsequently captured by CCD camera in order to make FRI images. FRI images underwent linear unmixing based on singular value decomposition (SVD) method in order to separate cellular from fluorescently labeled components. Non-negative matrix factorization (NMF) was also used to separate two quantum dot signals in FRI images.

Results

SVD & NMF algorithms have been written in MATLAB and applied to FRI images of phantom, containing two fluorophores, QD660 and QD710, in order to segregate fluorescent spectra. Peak signal to noise ratio (PSNR), signal to noise ratio (SNR) and contrast to noise ratio (CNR) for SVD & NMF methods.

Conclusions

Then used Statistical analysis based on one-way ANOVA and post-hoc analysis Results demonstrate that linear unmixing based on SVD could significantly

separate flourophores inferring optimal application for multispectral imaging in reflective mode.

Keywords: Linear Unmixing, Multispectral Imaging, Non-negative Matrix Factorization